

Customer Number

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:)	Group Art Unit: 1638
)	
St. Martin <i>et al.</i>)	Confirmation No.: 3349
)	
Application No.: 10/778,018)	Examiner: Keith O'Neal Robinson
)	
Filed: February 12, 2004)	Attorney Docket No.: 22727/04212
)	
For: Identification of Soybeans Having)	
Resistance to <i>Phytophthora Sojae</i>)	

Declaration of Dr. Brian W. Diers under 37 C.F.R § 1.132

I, Brian W. Diers, declare as follows:

1. I received a Ph.D. from Iowa State University and postdoctoral training at the University of Wisconsin. I have conducted research in the field of soybean breeding and genetics for 15 years. I have published some 55 full-length publications in peer-reviewed scientific journals.
2. Currently, I am a Professor of Breeding & Genetics at the Department of Crop Sciences, College of Agricultural, Consumer, and Environmental Sciences, University of Illinois at Urbana-Champaign.
3. I have directed research relating to marker assisted selection for yield, quality, and resistance to many pests and pathogens in soybean plants, including *Phytophthora sojae* for 15 years. I have published 12 research papers in this field, including: Diers *et al.*, 1992, Mapping *Phytophthora* resistance loci in soybean with restriction fragment polymorphism markers. Crop Sci. 32:377-383.

4. I am familiar with the work of Anne Dorrance and Steve St. Martin's group on the novel Rps8 trait locus.
5. I would like to comment on the process of marker assisted selection or MAS to clarify how scientists successfully perform MAS in a breeding program.
6. The process of MAS uses molecular markers that are good predictors of the presence or absence of a trait and is a well recognized industry approach that saves time and money in a breeding program. However, scientists in this field cannot use molecular markers to screen soybean populations for the presence of a novel trait or locus, such as the novel Rps8 resistance locus, unless they first discover a plant that carries that trait, map the locus or gene responsible for the trait, and finally "tag" the locus or gene to ascertain which molecular markers are associated with that gene.
7. A plant that carries a new Rps resistance trait is identified through inoculations with various *P. sojae* pathotypes. Once a plant with the desired trait is found, plant scientists need to map (i.e. discover the location of) the gene responsible for the trait. For example, before the work of the St. Martin group, seven *Rps* loci were identified for resistance to *P. sojae*. These *Rps* alleles were assigned to major linkage groups (MLG) as follows:

<u>Rps alleles</u>	<u>MLG</u>
<i>Rps1a, Rps1b, Rps1c, Rps1d, Rps1k</i>	N
<i>Rps2</i>	J
<i>Rps3a, Rps3b, Rps3c</i>	F
<i>Rps4</i>	G
<i>Rps5</i>	Tent G
<i>Rps6</i>	G
<i>Rps7</i>	N

Demirbas et al., 2001. Crop Sci. 41:1220-1227.

Diers et al., 1992. Crop Sci. 32:377-383.

This table shows that Rps resistance genes map to various MLG's. Therefore, when a new *Rps* locus is discovered, considerable experimentation is required to map the new *Rps* locus to a particular MLG and to a particular locus on the MLG. Once the *Rps* locus, and the particular molecular markers associated with that locus, have been

identified, those same molecular markers can be used for MAS of future progeny (from a parent that carries the *Rps* locus) to efficiently screen and select progeny segregating for the *Rps* locus in a faster, more efficient manner.

8. The *Rps8* resistance locus contains a novel gene that was not known before the work of St. Martin's group, published in K. D. Burnham, A. E. Dorrance*, D. M. Francis, R. J. Fioritto and S. K. St. Martin, *Rps8, A New Locus in Soybean for Resistance to Phytophthora sojae*, Crop Science 43:101-105 (2003). Plants that carry the *Rps8* trait locus (gene) were found to be resistant to combinations of *Phytophthora sojae* pathotypes vir1a, 1b, 1c, 1d, 1k, 2, 3a, 3b, 3c, 4, 5, 6 and 7. By contrast, plants that had all the previously identified *Rps* genes showed susceptibility to certain combinations of *P. sojae*. For example, a soybean plant that only carries the *Rps3* gene is susceptible to certain combination of *P. sojae* pathotypes, such as the combinations found in *P. sojae* race 17 (vir. 1b, 1d, 3a, 3b, 3c, 4, 5, 6, 7) and race 30 (vir 1a, 1b, 1k, 2, 3a, 4, 5, 6, 7). It is clear that a plant that only has the *Rps3* resistance locus does not display the phenotypic and genotypic resistance of a plant that carries the *Rps8* trait. Thus, an *Rps3* plant cannot be used to map and tag the novel *Rps8* trait locus. Only an *Rps8* carrying plant can be used for that purpose.
9. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

3-13-07
Date


Dr. Brian W. Diers